UKSAF Summer Meeting 2009

from Single Crystal to Wet Tissue: Thirty Years of UK Surface Analysis

Wednesday 1 July 2009

Host: University of Nottingham

Venue: Boots Science Building, University Park, Nottingham, NG7 2RD

09.30 - 10.00 Registration

10.00 - 10.10 Welcome & introduction
Dr Dave Sykes, UKSAF Chairman
Professor Saul Tendler, Head of the School of Pharmacy, University of Nottingham

10.10 – 10.55 Prof. John Vickerman, Manchester Interdisciplinary Biocentre, University of Manchester
Molecular SIMS – a Journey from Single Crystal to Biological Surface Studies

10.55 – 11.30 Prof. Neil Champness, Department of Chemistry, University of Nottingham
Surface Supramolecular Assembly

11.30 – 11.45 Coffee Break

11.45 – 12.20 Prof. Martyn Davies, School of Pharmacy, University of Nottingham
Challenges of Surface Characterisation in the Pharmaceutical Sector

(continued overleaf)

12.20 – 12.50 Dr Matthew Bunker, Molecular Profiles Limited, Nottingham

Surface Analysis of Pharmaceuticals and Biomaterials for Industry

12.50 – 13.00 **Annual General Meeting of the UK Surface Analysis Forum**

Agenda

1. Chairman's report
2. Honorary Treasurer's report & acceptance of the accounts for the year ending 31 March 2009
3. Honorary Secretary's report
4. Election of officers and members of the Executive Committee
5. Any other business

13.00 - 14.00 Lunch & Departmental Tours

14.00 – 14.30 Dr John Haycock, Department of Engineering Materials, The University of Sheffield

Creating Biological Structures and Function with Scaffolds and Surface Chemistry

14.30 – 15.00 Mr Yuan-Tsan Tseng, Department of Materials, University of Oxford

Biomaterials and Tissue Engineering

15.00 – 15.30 Ms Felicia Green, National Physical Laboratory, Teddington

Ambient Surface Mass Spectrometry: Characterisation of the Method DESI for Biomolecular Surfaces

15.30 – 16.05 Prof. Peter Weightman, Surface Science Research Centre, University of Liverpool

Surface Science under Water

16.05 – 16.30 Tea and Close of Meeting

Molecular SIMS – A Journey from Single Crystal to Biological Surface Studies

John C. Vickerman

Surface Analysis Research Centre,
Manchester Interdisciplinary Biocentre, The University of Manchester, Manchester, M1 7DN

A brief account will be given of journey made by the Manchester group and its collaborators in the development of molecular secondary ion mass spectrometry.

The earliest studies focussed on the application of static SIMS to single crystal surface studies. These studies successfully demonstrated that static SIMS delivered information on the delicate adsorbate state that mirrored that obtained by other surface science techniques. Subsequent application of the technique to studying the state and reactivity of bimetallic surfaces stimulated by collaboration with the Ertl group, demonstrated that static SIMS could be applied to the investigation of quite complex surface chemistry. This success stimulated the application of the technique to surface chemistry studies of much more complex systems such as polymers, ice mimics of polar stratospheric clouds, aerosols, culminating in biological systems.

The need to enhance ion yields of the larger biological molecules led to the development and introduction of polyatomic primary ion beams, most notably based on C_{60} buckminsterfullerene. This type of ion beam has transformed molecular analysis by SIMS. Not only have the yields of larger molecular ions been greatly increased, the bombardment induced damage that necessitated the static limit has been dramatically reduced such that for many materials the static limit requirement can be abandoned. A completely new analytical regime has opened up so that molecular depth profiling and 3D chemical imaging is possible. To fully realise the new capabilities for biological analysis a new generation of ToF-SIMS instrument is being developed that overcomes the compromises of pulsed beam instruments and that enables high mass resolution, high spatial resolution and high duty cycle to be attained simultaneously.

Surface Supramolecular Assembly

Neil R. Champness

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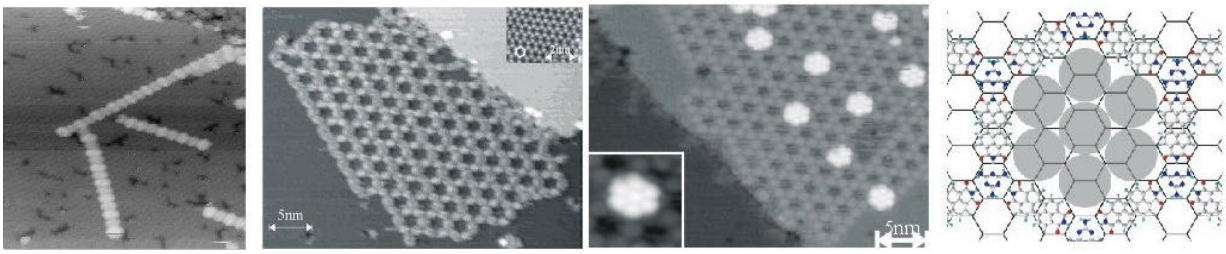
Non-covalent directional intermolecular interactions provide a pre-determined recognition pathway which has been widely exploited in supramolecular chemistry to form functional nanostructures in both solution

and in the solid-state. In this presentation recent advances in transferring the protocols of supramolecular organisation to two dimensional surface-based assembly will be discussed.[1]

We have demonstrated a major advance in this area – specifically the self assembly of an array of nanoscale pores which may be used as containment vessels. The presentation will detail the synthetic strategies used to construct surface frameworks and their use as templates for molecular entrapment and nanostructure formation. In particular an example of the formation of a bimolecular two-dimensional honeycomb array formed from melamine and perylenetetracarboxylic diimide (PTCDI) will be discussed (see Figure). This array, characterised by STM, accommodates pores that are 2.5nm in diameter, much larger than previously demonstrated, and have the capacity to accommodate several large molecules, for example C_{60} or C_{84}. The potential for wide application of this approach will be discussed with examples illustrating the effect of variation in molecular design. Most importantly our work establishes a direct connection between supramolecular chemistry and nanostructure fabrication.

More recent results have demonstrated protocols for studying such surface-based self-assembly using solution-phase deposition techniques.[3] Thus, we have demonstrated the role of guest species in the self-assembly process and the first direct imaging of a random molecular framework structure using rhombus-shaped molecules to give random rhombus tiling.



(i) (ii) (iii) (iv)

Figure: left to right: (i) formation of one dimensional chains of 1,4,5,8-naphthalenetetracarboxylic diimide (NTCDI) on a Ag/Si(111)-$\sqrt{3}$x$\sqrt{3}$R30^{o} substrate – chain length up to 20nm; (ii) multi-component (PTCDI-melamine) surface supramolecular framework on a Ag/Si(111)-$\sqrt{3}$x$\sqrt{3}$R30^{o} substrate – the structure forms large pores with a diameter of 3.4nm; (iii) use of pores as nanoscale containment vessels accommodating isolated heptameric C_{60} clusters; (iv) schematic representation of intermolecular interactions showing melamine vertices, PTCDI edges and captured C_{60} heptamers.

1. J.A. Theobald, N.S. Oxtoby, M.A. Phillips, N.R. Champness, P.H. Beton, *Nature*, 2003, **424**, 1029.

2. D.L. Keeling, N.S. Oxtoby, C. Wilson, M.J. Humphry, N.R. Champness, P.H. Beton, *Nano Lett.*, 2003, **3**, 9; J.A. Theobald, N.S. Oxtoby, N.R. Champness, P.H. Beton, T.J.S. Dennis, *Langmuir*, 2005, **21**, 2038; L.M.A. Perdigão, N.R. Champness, P.H. Beton, *Chem. Commun.*, 2006, 538; P.A. Staniec, L.M.A. Perdigão, B.L. Rogers, N.R.Champness, P.H. Beton, *J. Phys. Chem. C*, 2007, **111**, 886. L.M.A. Perdigão, A. Saywell, G.N. Fontes, P.A. Staniec, G. Goretzki, N.R. Champness, P.H. Beton, *Chem. Eur. J.* 2008*,* **14***,* 7600.

3. M.O. Blunt, J. Russell, M.C. Giménez-López, J.P. Garrahan, X. Lin, M. Schröder, N.R. Champness, P.H. Beton, *Science*, 2008, **322**, 1077; M. Blunt, X. Lin, M.C. Gimenez-Lopez, M. Schröder, N.R. Champness, P.H. Beton, *Chem Commun*., 2008, 2304.

Challenges of Surface Characterisation in the Pharmaceutical Sector

Martyn C. Davies

Laboratory of Biophysics and Surface Analysis, School of Pharmacy, University of Nottingham

Over the last few decades, there has been considerable activity in the search for novel pharmaceutical and biomedical systems to treat disease. Critical to the success of such systems, is the development of materials and devices that either deliver the active principle in a controlled fashion and/or promote appropriate cellular interactions with the local biological environment.

This talk will review the emergence of the role of surface tools for the study of the interfacial region of such systems in terms of their physicochemical properties and biological response, e.g., chemistry, surface energetics, topography and morphology. Examples discussed will be drawn from both conventional and advanced therapeutic systems and will range from the study of biomolecular interactions and gene therapy systems at the molecular level to the rapid high throughput screening of thousands of materials for optimum surface-cellular interactions.

Surface Analysis of Pharmaceuticals and Biomaterials for Industry

Matthew Bunker

Molecular Profiles Limited, Nottingham Business Park, NG8 6PX

Surface analysis lends itself to an incredibly diverse set of applications within the pharmaceutical, healthcare and biomedical fields. This talk will focus on disseminating case examples where surface analytical techniques have been used to understand material properties at the different stages of pharmaceutical development and helped to make selection decisions regarding process variables and manufacturing routes.

This is with respect to a wide variety of products ranging from drug delivery vehicles designed to release transforming growth factor-beta (TGF-b) proteins to vaccines to stents to raw excipients materials.
Examples will be shown where surface analytical techniques have been used both proactively and reactively to help better understand the mechanisms of drug release, optimise biomolecule loading and correlate relationships between material solid state properties and processing with a view to predicting subsequent behaviour.

The important role surface analytical techniques have in ensuring the design and manufacture of a drug product in alignment with the most recent regulatory guidance will be covered. In addition the future that surface science holds for helping develop formulations designed to deliver poorly water soluble drugs will be highlighted.

Creating Biological Structures and Function with Scaffolds and Surface Chemistry

John W. Haycock

Kroto Research Institute, Engineering Materials, University of Sheffield, Broad Lane, S3 7HQ

This talk will focus on the design of scaffolds and bioengineering approaches for repairing peripheral nervous system injuries. It will introduce scaffold fabrication using hydrolysable microfiber polymers and then consider modifying the surface chemistry of these scaffolds for optimizing cell growth and survival. It will then consider creating bioactive surfaces for generating specialized properties e.g. anti-inflammatory biomaterials.

Peripheral nerve injuries are common and although regeneration is possible axon damage is often to significant. Generally, surgical intervention is needed in order for any functional sensation to be regained. Although the gold standard for peripheral nerve repair is autografting there are major disadvantages here, including a lack of donor nerve or donor site morbidity. The high level of cell death and lack of coherent orientation of regenerating axons without surgical intervention has lead to therapies using nerve guidance conduits (NGCs). In the simplest form NGCs are hollow tubes that act as a physical guide between the proximal and distal stumps, bridging the gap of the injury. NGCs can be made from a variety of synthetic or natural materials and incorporate cells and growth factors to improve guidance. The aim of our present work is to improve the NGC design by placing parallel degradable microfibres within an NGC. Fibres are intended to support Schwann cell growth initially *in vitro* and in turn encourage more complete axon alignment within the device following implantation.

In order to optimize this we rely on the surface chemical modification of microfibers by plasma polymerization, and fabricate 3D scaffolds by culturing Schwann cells within a closed bioreactor. Our data shows that PLLA fibres will readily sustain Schwann cell growth, but that cellular viability is only about 60% after 96 hours in culture. However, this increases significantly to 90% when the PLLA fibres are modified using a plasma polymerized layer of acrylic acid. The total number of adhered Schwann cells per microfiber is also increased under these conditions.

Thus the value of surface modification of biomaterials is extremely important for directing cell function, and this can be extended to creating bioactive surfaces with specialized properties e.g. anti-inflammatory biomaterials. We have therefore devised a method for the rapid fabrication of anti-inflammatory biomaterials based on the synthesis of a resorcinarene-MSH peptide conjugate. The resorcinarene has a high affinity for a wide range of material surfaces and adheres by many weak non-covalent interactions. The MSH peptide motif itself contains anti-inflammatory activity, such that when nerve and skin cells are grown on these surfaces they display a reduced response to proinflammatory signals. This work therefore has potential value in the surface modification of implantable materials which are associated with causing an inflammatory response and in turn the failure of the device.

Biomaterials and Tissue Engineering

Yuan-Tsan, Tseng

Department of Materials, University of Oxford, UK

Biomaterials and their applications are having a tremendous impact on health care technology. For example, the prosthetics that are used in hip replacements, arterial stents and heart valves are providing significant improvements to the quality of life of patients.

The next challenge is developing medical devices with regenerative capability- tissue engineering. Collagen is the most abundant ECM protein, which is used to manufacture scaffolds as a matrix for tissue engineering. Indirect solid freeform fabrication was developed to manipulate the collagen matrix into the desired shape. In addition, controls of the collagen scaffolds along several length scales was achieved:

1. at the molecular level in stochemistry and substitution level of Hydroxyapaptite
2. at the micro-structural level in volume fraction of Hydroxyapaptite, porosity of collagen and micro-channel to aid vascularisation
3. Macroscopic level - to fit the matrix into the proposed defect site.

Another aspect of the biomaterials involves the development of biodegradable local drug delivery devices. A higher drug concentration in relevant tissue administrated by a local drug delivery device should reduce any systemic toxicities and side effect compared to parental administration. With the potential in bone local drug delivery application a novel poly(lactic-co-glycolic acid) (PLGA) microsphere coated with HA to produce a local drug delivery device with osteoconductivity.

Ambient Surface Mass Spectrometry: Characterisation of Desorption Electrospray Ionisation (DESI) Mass Spectrometry for Biomolecular Surfaces

Felicia M. Green[1],
T. L. Salter[1], D. G. Castner[2], L. J. Gamble[2], I. S. Gilmore[1], P. Stokes[3], G. O'Connor[3]

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The surface of a biomaterial is the interface between that material and the biological environment. The surface structure and composition of a biomaterial mediates the reactions that occur when it is placed into the body. Biomolecular engineering is key to achieving the desired functionality. Many surface

analytical techniques used to study these systems are vacuum techniques (e.g. SIMS, XPS). Whilst these techniques are very powerful the requirement for vacuum is a major limitation for the analysis of delicate biological samples. Desorption Electrospray Ionisation (DESI) shows great potential to give ambient surface mass spectrometry of these systems. Here, we study the dependency of surface chemistry on the desorption efficiency to help improve understanding of the DESI surface sensitivity and desorption mechanism.

By studying two model systems, we were able to explore different factors affecting biomolecular desorption using DESI. The first investigates the desorption efficiency of DESI when changing the surface interaction. Here, a model system of phenylalanine was used, evaporated on to a range of surfaces, glass, uv/ozone treated glass and ptfe with a range of wettabilities. Three different electrospray solvent compositions MeOH:H_2O, ACN:H_2O and Toluene:H_2O with varying fractions of organic solvent were used. These showed a greater than 0.5 solvent fraction gave better spatial resolution, with a 0.9 fraction of organic solvent (ACN or MeOH) giving a spot size of a factor 2 smaller. In parallel, we see an exponential increase in molecular signal intensity with increased solvent fraction (ACN or MeOH) leading to significantly better desorption efficiency. The effect of the substrate surface on desorption efficiency can be related to wettability, PTFE (low wettability) is a factor of 10 lower at optimum settings compared with glass and UV/ozone cleaned glass (high wettability). This study shows, that the parameters contact angle and droplet size are more important than solubility for improving DESI desorption efficiency.

We developed a systematic study to understand the efficiency of DESI for biomolecule desorption, by producing a set of well controlled SAMS surfaces with different binding strengths to an attached peptide. Through comparison of peptide molecular ion signal from DESI and SIMS, desorption efficiency of DESI can be related to the strength of surface binding of the peptide. Here, we show that DESI can only detect unbound or loosely interacting molecules. These systems allow the different chemical, surface and ionisation mechanisms occurring during DESI desorption to be separated out and understood individually, to enhance our knowledge and optimisation of the DESI desorption efficiency.

Surface Science Under Water

Peter Weightman

Physics Department and Surface Science Research Centre, University of Liverpool

The development of ultra high vacuum (UHV) techniques in the 1960's made experimental surface science possible. Since that time we have made major advances in the understanding of surfaces and surface processes through the use of a large variety of UHV experimental techniques. However there has always been a major weakness in our approach to the study of surfaces in that very few of our

experimental probes can operate in ambient conditions. This is a particular problem in the study of biological systems that require a liquid environment in order to exhibit their functional behaviour.

Fortunately the last decade has seen the development of a number of techniques that can be used to study surfaces under liquids. This talk will describe the results of research with one of these techniques that has already shown its potential for studies of the solid/liquid interface, reflection anisotropy spectroscopy, and outline technological advances in the development of a second technique, terahertz spectroscopy, for which operating in water is a particularly difficult challenge.